Interaction studies between these two peptides are currently in progress and will allow us to define the complex structure as well as the amino acids involved in the interaction.

2076-Pos Board B62
Structural Studies of Super-Mini-B: Role of the N-Terminal Insertion Sequence of Surfactant Protein B
Mahzad Sharifahmadian, Alan J. Waring, Frans J. Walther, Rober H. Notter, Valerie Booth.
Lung surfactant is a mixture of phospholipids and proteins that is critical for breathing. The hydrophobic surfactant protein B (SP-B) is essential for the function of lung surfactant. Super Mini-B is a 41-residue peptide with internal disulfide bonds that contains the N-terminal 7-residue insertion sequence and the N-and C-terminal helices of SP-B. It has previously been shown to retain the similar activity to full-length SP-B as measured by dynamic compliance and arterial oxygenation in rat studies. Interestingly, Super-Mini-B performs better than peptides which lack the N-terminal insertion sequence (e.g. Mini-B). We have used circular dichroism and solution and solid state NMR to study Super-Mini-B's structure and lipid interactions in various environments, including anionic micelles. Comparison of the results for Mini-B and Super Mini-B help to unveil the contribution of the 7-residue insertion sequence to the function of SP-B.

2077-Pos Board B63
Investigating the Relationship Between Physical Properties of Detergents and Membrane Protein Structure Determination
Ryan Oliver, Ryan Lo, Justin Kim, Linda Columbus.
Membrane proteins comprise only 1.8% of the atomic-resolution structures deposited in the RSB Protein Data Bank (http://pdbtm.enzim.hu). One bottleneck contributing to the under-representation of membrane protein structures is the requirement of a membrane mimic, such as a detergent micelle, to solubilize the membrane protein and form a stable protein-detergent complex (PDC). The mimic that leads to a structure is typically determined by empirically and exhaustively screening commercially available detergents. Understanding the relationship between detergent micelle physical properties and the stability, fold, and function of membrane proteins in PDC's reduces the extent of empirical screening required. Additionally, many structures are determined in detergent mixtures and with additives that affect the micelle properties and the protein-detergent complex. The aim of this study is to investigate micelle physical properties, both pure and mixed, and with additives commonly used in structural biology such as polyethylene glycols and cholesterol. In particular, we will present the thermodynamic properties of micelle formation (using isothermal titration calorimetry) and micelle size and shape (using small angle x-ray scattering) of binary detergent mixtures, as well as with additives. We will also present preliminary NMR data of membrane proteins in the characterized detergent micelles in order to investigate trends of detergent properties that correlate with stabilization of a protein fold. These results provide a foundation for understanding the role of detergents in membrane protein structure determination.

2078-Pos Board B64
NMR Backbone Assignment of Opai: A Mediator of Host:Neisseria Interactions
Daniel A. Fox, Linda Columbus.
Opa (opacity associated) proteins from Neisseria gonorrhoeae (NG) and Neisseria meningitides (NM) induce phagocytosis of the bacterium by non-phagocytic cells such as epithelial cells. There are eleven NG and four NM Opa alleles that undergo phase-variation expression to the outer membrane. The protein products are nearly identical in sequence, but vary in three extracellular loop regions that determine the host receptor specificity. Opa proteins bind to carcinoembryonic antigen-like cellular adhesion molecules (CEACAMs) or to heparansulfate proteoglycan receptors (HSPGs), thus subdividing Opa proteins into two classes, OpaCEA and OpaHS, respectively. Mutational and chimeric experiments have not revealed the sequence determinants of the hypervariable regions that are responsible for receptor recognition. The goal of this study is to investigate the structure, dynamics, and receptor interactions of OpaI, a 27 kDa, 238 amino acid OpaCEA from NG MS11. OpaI was over-expressed in E. coli, purified, and refolded in dodecylphosphocholine. Two-dimensional 15N, 1H- TROSY spectra, CD, and SDS-PAGE gel mobility of the protein-detergent complex indicate that the protein is folded and well-suited for NMR studies Using TROSY-based pulse sequences, methyl labeling, and specific amino acid labeling, a suitable backbone assignment was achieved and an initial low-resolution structural calculation with H-bond and NOE-derived restraints is presented.

2079-Pos Board B65
Investigation of the Neisseria Gonorrhoeae Opacity Associated Membrane Protein a Structure, Dynamics, and Host-Receptor Interactions
Ryan H. Lo.
Opacity associated (Opa) outer membrane proteins from the bacteria *Neisseria meningitides* and *Neisseria gonorrhoeae* bind to human host cell receptors, which triggers phagocytosis of the bacteria. Opa proteins are classified into two groups (Opa_{CEA} and Opa_{HS}) based on host receptor specificity. Opa_{CEA} interacts with carcinoembryonic antigen-related cell adhesion molecules (CEACAM) and Opa_{HS} binds heparansulfate proteoglycan (HSPG) receptors. Two hypervariable extracellular loops are proposed to determine receptor specificity; however, the molecular determinants of the interaction and host receptor specificity are unknown. The aim of this study is to determine the structure of OpaA (an Opa_{HS} from *N. gonorrhoeae* MS11) using NMR, and to map the specific loop interactions of OpaA with heparin, a competitive inhibitor of the Opa_{HS} and HSPG interaction. The expression, purification, and folding of OpaA will be presented as well as the progress towards the NMR backbone assignment. In addition, the feasibility of using chemical shift perturbation to map the OpaA - heparin interaction will be explored. These invesitgations of the binding specificity of OpaA to heparin will further the understanding of the pathogenesis of *Neisseria*, which will in turn increase our understanding of an attractive function for receptor-targeted therapeutics.

2080-Pos Board B66
NMR Structural Studies of a Two-Transmembrane Helix Segment of the Na+/H+ Exchanger Isoform 1
Brian L. Lee, Claudia Alves, Brian D. Sykes, Larry Fliegel.
The Na^+/H^+ exchanger isoform 1 (NHE1) is an integral membrane protein important in the regulation of intracellular pH in the heart by exchanging one intracellular H^+ for one extracellular Na^+. It contains an N-terminal transmembrane (TM) domain responsible for ion transport and a C-terminal, cytoplasmic, regulatory domain.
There is currently no high-resolution structure of NHE1; however, a topology model of the protein by Wakabayashi *et al*. (*J. Biol. Chem.***275,** 7942) suggests that the membrane domain (amino acids 1-500) contains 12 TM helices. We have previously used NMR spectroscopy to determine the structures of peptides representing several TM helices from NHE1. Structures have been determined for TM IV in organic tri-solvent mixture, and for TM helices VII, IX, XI, and VI in DPC micelles.
To provide further insight into the three-dimensional structure of NHE1, we are attempting to express and determine the structures of multiple TM helices. We have recently expressed and purified a two-TM helix segment representing TM VI-VII (amino acids 226-274) of NHE1. These two TM helices have been previously shown to be important in NHE1 function. Furthermore, modelling of NHE1 by Landau *et al.* (*J. Biol. Chem.* **282,** 37854) suggest that these two helices interact and play key roles in ion transport. We are determining the structure of this region in micelles using NMR. NMR spectra of unlabelled TM VI-VII peptide show good quality spectra for structure determination. Further experiments with ^{15}N labelled peptide will further define the structure. This will provide insight into TM helix interactions in NHE1 and help us to build up the structure of the protein from these TM segments. (Supported by AHFMR, HSF, CIHR, and AIHS).

2081-Pos Board B67
Investigation of the Three-Dimensional Structure of the Caveolin-1 membrane Interacting Domain
Jinwoo Lee.
The importance of understanding membrane protein structure and function is growing after scientists have recognized that approximately one third of the genome encodes for membrane proteins. Caveolin is an integral membrane protein that is found in high abundance in caveolae. Caveolae are invaginations of the plasma membrane that play an important role in many cellular functions. Interestingly, caveolin plays a dual-role in caveolae. On one hand it is responsible for forming the ''flask-like'' shape of caveolae, and on the other hand it is intimately involved in cellular processes such as signal transduction and protein trafficking. Alterations in the levels of caveolin protein and mutations of the caveolin protein have been implicated in a variety of disease states including cancer, heart disease, and muscular dystrophy.
Caveolin possesses a unique hairpin-like topology (N- and C- termini are on the same side of the membrane) in the membrane which is thought to facilitate its functions. In particular, mutations of two proline residues (P110A and P132L) in the caveolin-1 transmembrane domain appear to